



Betty Ford Center

Recovery.
State of the art treatment center
dedicated to their patients rehab.

Get Help Today
(800) 392-7540
bettyfordcenter.org



October 2, 2011

Reformulating Injectables for Oral Delivery

By Erik Greb

Could oral absorption-enhancing technologies change the shape of protein delivery?

The oral delivery of biopharmaceuticals is highly desirable because it would provide ease of administration and improve patient compliance. But oral administration remains an elusive goal in biopharmaceutical drug delivery. To date, major obstacles have included problems of bioavailability caused by low absorption, poor cellular permeability, and protein instability and degradation. Several specialty pharmaceutical and other companies are applying various absorption-enhancing technologies to address the problem with some success. Nevertheless, challenges remain, including limitations on the size of the macromolecules that can be delivered.

The challenges



Formulation concerns. Achieving sufficient absorption of proteins is the most crucial challenge in formulation development, according to Wei-Guo Dai, a research fellow in drug delivery and device development at Johnson & Johnson Pharmaceutical R&D. Delivering an adequate amount of protein into circulation by oral delivery is difficult because enzymes in the body degrade the protein, the gastrointestinal membrane is poorly permeable, and large molecules cannot easily enter into the bloodstream intact. Past efforts involving particulate delivery systems (e.g., nanoparticles or liposomes, multifunctional polymers, and enzyme inhibitors) have achieved low rates of absorption that were not adequate to achieve therapeutic efficacy, says Dai.

ILLUSTRATION: MELISSA MCEVOY. IMAGES: JEAN-PAUL NACIVET/GETTY IMAGES; TOM NULENS

Permeation enhancers are one strategy to improve oral absorption. These agents include surfactants, fatty acids, and bile salts. They can be added to the formulation as excipients, or incorporated into the delivery systems using proprietary technologies. Permeation enhancers function by disrupting the intestine's epithelial membrane or by loosening the tight junctions (i.e., intracellular barriers) between epithelial cells.

In certain cases, permeation enhancers have enabled formulators to achieve 10% bioavailability in preclinical research studies. But the amount of permeation enhancers required in these studies raises concerns about toxicity. Various types of permeation enhancers entail various levels of risk, but even agents that are generally recognized as safe, such as ethanol and fatty acids, could pose toxicity problems because of the dosage amount and frequency. Formulators thus must conduct comprehensive toxicity studies, says Dai.

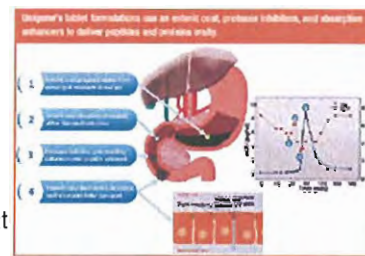
Another strategy to compensate for low bioavailability is to increase the dose of protein in a tablet. For example, if a tablet contained hundreds of milligrams of insulin—far more than is administered in an injection—and if 1–5% of it were absorbed into the bloodstream, it could be enough to achieve the desired therapeutic effect. But this approach has limitations. "You may be able to jack up the dose, but your absorption window may be so small that you just don't have enough time for the drug to be absorbed," says Carlos N. Velez, managing partner of LacertaBio, a biotechnology business-development consulting firm.

Aside from absorption issues, increasing the level of protein in an oral formulation may not be cost-effective in the absence of a superior clinical benefit. "My guess is there aren't enough suppliers for some of these proteins to bring the cost down to the point where you've got a \$5-per-day dose," says Velez. He cites one company that has an oral formulation of a modestly sized peptide, but cannot buy the active ingredient at a low price. "It'll be an enormous

dollar-per-day dose, and that program is probably not going to get very far because of a supply-cost issue," says Velez.

Manufacturing obstacles. In addition to formulation challenges, manufacturing a protein-based tablet would entail many production difficulties. "The way that tablets and capsules are made today is not at all conducive to making a protein product," says Mike Pikal, Pfizer distinguished chair in pharmaceutical technology and professor of pharmaceuticals at the University of Connecticut.

One major problem is that proteins must be incorporated into a matrix, typically a disaccharide. Most tablet-production facilities run at a controlled humidity of 40%, but because the disaccharide matrix is extremely hygroscopic, the formulation becomes a paste under those conditions, "and it's a mess," says Pikal. Operating a plant at 2% humidity would avoid this problem, but this solution could be impractical because the technology would be extremely expensive for a tablet manufacturing facility.



Unigene's tablet formulations use an enteric coat, protease inhibitors, and absorption enhancers to deliver peptides and proteins orally. (IMAGE IS COURTESY OF UNIGENE LABORATORIES)

Another strategy might be to enclose much of the production line with barrier technology to keep oxygen and moisture out until the tablets are packaged. "Using the best of the techniques that are available, you can probably do that, but it's a significant engineering challenge," says Pikal. "While barrier systems are becoming common in parenteral operations, as far as I know, such systems are not available for tablet operations," he adds. It's also conceivable that a manufacturer would have to make major changes to the air-handling and humidity-control systems for its plant.

In addition, amorphous disaccharide matrices that contain proteins are not particularly compressible, so formulators would have to add ingredients (e.g., diluents or disintegrants) to achieve a mixture that could be made into a tablet more readily. Also, it seems unlikely that a protein formulation could be wet granulated, so processing methodology would be limited, says Pikal. The amount of excipients required likely would limit the amount of protein that the tablet could contain. As a result, the amount of protein in the tablet "is going to have to be awfully small," according to Pikal, and manufacturers might be limited to working only with potent proteins for their tablet.

Besides causing problems for the manufacturing process, moisture also can degrade the protein itself. "In some cases, you can break just one of those amino acid or disulfide bonds, and the activity of the molecule is destroyed," says Velez. A traditional wet granulator and tablet punch may not be peptide-friendly, he adds. A tableting method would need to ensure protein stability and activity throughout the process. This tableting process would have to include analytical methods to monitor the protein's structure. Personnel possibly could extract sample tablets during the process, for example, then extract the protein and analyze it with X-ray crystallography.

Oral absorption-enhancing technologies

Oral absorption-enhancing technologies are one way to address the problem of poor bioavailability encountered in biopharmaceutical drug delivery. Several specialty pharmaceutical companies have developed such technology platforms.

Passive transcellular transport through small-molecule carriers. One approach taken by the biopharmaceutical company Emisphere Technologies uses small-molecule carriers to enable the passive transcellular transport of drug molecules across cell membranes. The company has developed a library of about 4000 proprietary small-molecule compounds, including acylated amino acids. The compounds' molecular weights typically are less than 400 Da and are the basis of Emisphere's Eligen Technology.

The company uses a screening process to match the properties of its carriers (e.g., solubility) to those of the drug to be formulated. The goal is to find carriers that will help protect the drug and interact with it in the right way to deliver it into circulation.

The carriers and the protein are blended together in a conventional process, but manufacturers must control parameters such as heat to ensure that the protein is not damaged. "It's really, in most cases, blending the carriers and proceeding with the process as you would with any other oral drug," says Prateek Bhargava, Emisphere's manager of formulations. The carriers can be used in most processes (e.g., milling, blending, and compression) and require no special handling.

After blending, the carrier and protein engage in a weak, noncovalent interaction that does not alter the protein's form. The process is similar to that of physically mixing a small-molecule active ingredient and excipient. This interaction protects the protein from rapid degradation as it enters the acidic environment of the stomach and confronts enzymes in the small intestine. The carriers transport the therapeutic molecule through the gastrointestinal tract quickly, which also reduces the opportunity for protein degradation, says David Gschneidner, Emisphere's director of chemistry.

Unlike other carrier-based drug-delivery systems that typically use permeation enhancers, the Eligen Technology conveys its payload through passive transcellular transport. The carriers cause a transient fluidization of the membranes in the gastrointestinal tract, thus increasing their permeability. After the membrane absorbs the

formulation, it closes again. "Typically, we start to see absorption in the stomach. It can also continue into the upper intestine, all the way through the small intestine, but we typically see a rapid uptake of the drug, says Gschneidner.

After the formulation enters the bloodstream, it is greatly diluted. The large volume of blood is enough to break the weak noncovalent bond between the carrier and the protein, thus causing the two to separate in a passive process. The carrier is eliminated through normal excretory processes.

The largest molecule that the Eligen Technology has delivered is human growth hormone, which has a molecular weight of about 22 kDa. The carriers also have been able to deliver heparin, which has a molecular weight of 12–15 kDa, and insulin, which has a molecular weight of almost 6 kDa.

Emisphere has not yet marketed an oral formulation of a large molecule, but has one product, salmon calcitonin, a 32-amino-acid linear polypeptide hormone, in Phase III clinical trials. The drug, a treatment for osteoporosis and osteoarthritis, is being developed jointly with Novartis. In addition, Emisphere is collaborating with Novo Nordisk on the oral delivery of glucagon-like peptide-1 (GLP-1) and insulin analogs. The GLP-1 analogs are in Phase I trials, and the oral insulin is in preclinical development.

Enterically coated tablets. To deliver biopharmaceuticals orally, Unigene Laboratories creates enterically coated tablets that include permeation enhancers and protease inhibitors, which prevent protein degradation. The protease inhibitor is citric acid, an organic acid that, when released from the tablet, creates a transient, localized, acidic microenvironment in which enzymes cannot readily break down the peptide.

The permeation enhancer is an acyl carnitine, L-lauroyl carnitine. By affecting calcium transport and levels of adenosine triphosphate within the epithelial cells of the intestine, the acyl carnitine temporarily loosens the tight junctions between these cells. This action frees the peptide to pass through these junctions by a mechanism called paracellular transport.

The tablet formulation is protected by a pH-sensitive, poly(meth)acrylate based enteric coat that remains stable in the acidic environment of the stomach. The coating, made from Evonik's Eudragit polymers, keeps the tablet intact and prevents the peptide from being absorbed in the stomach. When the tablet enters the duodenum, which is a neutral environment, the enteric coating dissolves. The location of drug release can be adjusted by using an Eudragit polymer of the appropriate pH. Altering the thickness of the enteric coating affects the timing of drug release and can help obtain the best bioavailability and consistency.

A water-soluble subcoat ensures that the enteric coating dissolves completely before the tablet releases its acid (i.e., the protease inhibitor) into the intestine. Otherwise, the release of the acid could prevent the rapid and complete dissolution of the enteric coat. The subcoat thus ensures that the formulation "is much more consistent in its release characteristics than it would have otherwise been," says Nozer Mehta, Unigene's vice-president of biological R&D.

Paracellular transport raises the risk that unwanted materials, such as other small, digested food-based peptides, may pass through the tight junctions after they have been opened. This risk is mitigated because the enteric coating is designed to remain intact until the tablet opens in a small area in the duodenum. The coating thus prevents acyl carnitine from opening tight junctions throughout the gastrointestinal tract. Unigene's studies also show that the tight-junction opening is a transient phenomenon (1). "If you gave the permeation enhancer 30 minutes in advance and then gave the peptide, you would not see any absorption, says Mehta.

Paracellular transport does limit the size of the molecules that can be delivered. Unigene has delivered insulin successfully, and it generally focuses on molecules made of fewer than 50–60 amino acids. "Large proteins probably would not work with our technology," says Mehta.

Peptides do not have to be derivatized or changed to be compatible with Unigene's oral technology. In addition, the method requires no special manufacturing equipment other than spray coaters to apply the subcoat and the enteric coat to the tablet.

Unigene and Tarsa Therapeutics recently completed a Phase III study of a calcitonin formulation made with Unigene's oral delivery technology. Unigene also is using the technology to develop a parathyroid hormone analog, currently in Phase II development, with GlaxoSmithKline.

Other commercial approaches

Various other companies have developed their own approaches to delivering large molecules orally. Merriam Pharmaceuticals's GIPET technology platform uses small- and large-molecule surface-active materials to enhance drug absorption in the small intestine. The surface-active agents used in GIPET are generally regarded as safe (GRAS) materials, including fatty acids and derivatives, surfactants (e.g., mono- or diglycerides), and lecithin.

GIPET materials form mixed-micelle vesicular structures and liquid crystals that entrap the active compound through physical or physical-chemical interactions, according to the company. Mixing the enhancer and drug sometimes

increases the latter's lipophilic properties, thus enabling large molecules to cross the gastrointestinal membrane. The micelles and liquid crystals also protect drug molecules from enzymatic degradation.

By acting as mild surfactants, GIPET enhancers increase the fluidity of the apical membrane and aid the transcellular transport of drug molecules. The agents also can increase the paracellular transport of drugs by contracting cytoskeletal actin filaments, which open the tight junctions and permit increased permeation of hydrophilic compounds. This function is not considered a significant component of the activity of GIPET, however.

The technology can be used to create three types of dosage forms. GIPET I yields enteric-coated tablets that contain the surface-active materials in powder form, along with the drug. GIPET II creates microemulsions of oil, surfactant, and drug in an enteric-coated gel capsule. GIPET III produces a mixture of fatty-acid derivatives in an enteric-coated gel capsule. All three GIPETs can deliver peptide or protein drugs. GIPET dosage forms can be created using conventional solid oral dosage manufacturing equipment.

In Phase I clinical studies, GIPET successfully delivered acyline, a decapeptide that can be used to treat prostate and breast cancer, by the oral route and achieved pharmacological activity. GIPET also delivered a peptide of a similar size, desmopressin, in the clinic. Merrion currently is collaborating with a partner to deliver larger peptides with GIPET. No products created through this technology have been marketed, however (2).

Intravail technology from Aegis Therapeutics uses transmucosal absorption enhancers for the oral delivery of potent peptides and proteins. The Intravail absorption enhancers comprise a group of alkylsaccharides related to mild surfactants common in personal care and food products. The GRAS enhancers do not irritate mucosal membranes.

Studies conducted at Albany Medical College have shown that the Intravail excipients can promote the oral absorption of certain peptides, such as octreotide and the antiobesity-antidiabetic leptin fragment D-Leu-OB3. The technology provides bioavailability comparable to or exceeding that achieved by subcutaneous injection. In one study, Intravail increased the oral absorption of an antibreast-cancer peptide by a factor of 10 and reduced the projected daily oral human dose from 2 mg/day to 200 µg/day. In addition to increasing bioavailability, the Intravail excipients have been shown to prevent peptide and protein aggregation and reduce resulting immunogenicity.

The Intravail technology delivers protein therapeutics as large as 30,000 Da, including GLP-1 analogs, calcitonin, growth hormone, leptin, insulin, erythropoietin, and low molecular weight heparins. Intravail formulations are compatible with conventional manufacturing processes for flash-dissolve wafers, tablets, and gelcaps.

In August 2011, Aegis received a patent for its Intravail-based oral formulations of several GLP-1 analogs. In April 2011, Aegis and scientists at Albany Medical College found that an Intravail-based oral formulation of the peptide octreotide achieved systemic bioavailability that exceeded that achieved by subcutaneous injection.

The NanoCrystal technology from Alkermes (formerly Elan Drug Technologies) is designed to increase drugs' solubility and can be used to deliver proteins and peptides (3). First, nanosized drug particles are created through methods such as wet milling, homogenization, precipitation, and supercritical-fluid techniques. This step increases the drug's surface area and enhances its dissolution profile. Proprietary stabilizers, such as F68 and sodium deoxycholate, are then surface adsorbed onto the drug particles to prevent agglomeration. This process yields an aqueous dispersion that behaves like a solution and can be sprayed onto a substrate, such as lactose, and compressed into a tablet (4).

Scientists have used the NanoCrystal technology to deliver zinc insulin orally. By altering peptides' particle size and surface properties, NanoCrystal technology can improve bioavailability by as much as 600%, according to the company. The technology also enabled sirolimus, an injectable cancer compound sold by Pfizer, to be formulated as an oral tablet. The technology can boost the drug's rate of absorption and reduce the size of the required dose.

Researchers' solutions

Researchers at universities also are looking for methods of delivering peptides and proteins orally. A research team at Kyoto Pharmaceutical University created a gastrointestinal mucoadhesive patch system (GI-MAPS) consisting of four layered films contained within an enteric capsule. The system's backing layer is made of ethyl cellulose (EC), a water-insoluble polymer. Its surface layer is made of an enteric pH-sensitive polymer (e.g., hydroxypropylmethylcellulose phthalate) coated with an adhesive layer. The middle layer, made of cellulose membrane, contains the drug and permeation enhancers (e.g., organic acids and surfactants), and is attached to the backing layer by a heating-press method. The surface layer is attached to the middle layer with an adhesive layer made of carboxyvinyl polymer.

After oral administration, the system's surface layer dissolves and adheres to the mucosal membrane of the small intestine. This result creates a closed space that contains the drug and the absorption enhancer. The high-concentration gradient in the closed space causes the small intestine to absorb the protein through passive diffusion.

The GI-MAPS enabled the researchers to deliver recombinant human granulocyte colony-stimulating factor orally to dogs and obtain bioavailability of 23%. By collaborating with the semiconductor industry, drug-delivery company BioSerenTach developed a machine that produces GI-MAPS under GMP conditions (5, 6).

Particulate systems, including solid lipid nanoparticles (SLN), have been investigated as carriers for peptides, proteins, and antigens. Scientists have incorporated proteins and antigens into SLN and administered them orally. SLN formulations have improved protein stability, protected proteins from degradation, and achieved sustained drug release. Cyclosporine A, insulin, calcitonin, and somatostatin have been incorporated into solid lipid particles and are currently being studied (7).

Researchers at St. John's University in Queens, New York, formulated oral dosage forms of β -lactamase (BLM) within self-nanoemulsifying drug-delivery systems (SNEDDS). The scientists composed a SNEDDS of propylene glycol monolaurate, polyethoxylated castor oil, and 2-(2-ethoxyethoxy)ethanol. They loaded approximately 2200 mU of BLM into the oil phase of SNEDDS using a solid-dispersion technique (8, 9).

In vitro tests of transporting BLM across a cell monolayer showed that the SNEDDS resulted in 33% cumulative transport of BLM at 5 h while the BLM free solution achieved negligible transport. Oral delivery of 4500 mU/kg of BLM in a SNEDDS to rats achieved a bioavailability of 6.34%, which was 1.5 times greater than that achieved by BLM free solution. Delivery of BLM in the aqueous phase of the nanoemulsion resulted in a pharmacokinetic profile similar to that obtained by the free solution (10). The scientists currently are continuing these studies.

Conclusion

Scientists have made progress in developing tablet formulations that can protect proteins from the digestive system and deliver them into the bloodstream. The level of bioavailability that current formulations achieve may not be adequate for clinical or commercial purposes, however. Even if these problems were to be solved, manufacturers would still need to develop new manufacturing and analytical methods that safeguarded proteins' stability and ensured their biological activity.

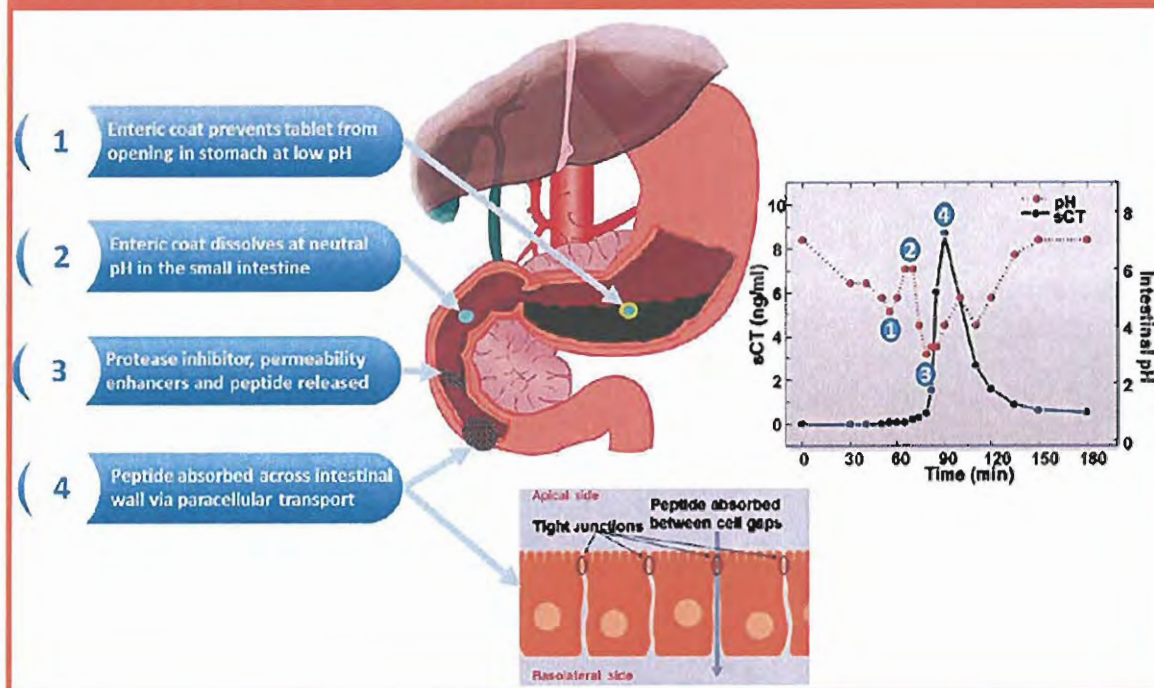
References

1. J.H. Hochman, J.A. Fix, and E.L. LeCluyse, *J. Pharm. Exp. Therap.* **269** (2), 813–822 (1994).
2. E.G. Walsh, J.S. Fox, and T.W. Leonard, *Pharm. Technol.* **35** (4), s12–s14 (2011).
3. E. Merisko-Liversidge, S.L. McGurk, and G.G. Liversidge, *Pharm. Res.* **21** (9), 1545–1553 (2004).
4. C. Timpe, *Am. Pharm. Rev.* **11** (1), 12–21 (2010).
5. S. Eiamtrakarn et al., *Biomater.* **23** (1), 145–152 (2002).
6. J. Shaji and V. Patole, *Indian J. Pharm. Sci.* **70** (3), 269–277 (2008).
7. A.J. Almeida and E. Souto, *Adv. Drug Deliv. Rev.* **59** (6), 478–490 (2007).
8. S.V.R. Rao and J. Shao, *Int. J. Pharm.* **362** (1–2), 2–9 (2008).
9. S.V.R. Rao, P. Agarwal, and J. Shao, *Int. J. Pharm.* **362** (1–2), 10–15 (2008).
10. S.V.R. Rao, K. Yajurvedi, and J. Shao, *Int. J. Pharm.* **362** (1–2), 16–19 (2008).



ILLUSTRATION: MELISSA MCEVOY. IMAGES: JEAN-PAUL NACIVET/GETTY IMAGES; TOM NULENS

Unigene's tablet formulations use an enteric coat, protease inhibitors, and absorption enhancers to deliver peptides and proteins orally.



Unigene's tablet formulations use an enteric coat, protease inhibitors, and absorption enhancers to deliver peptides and proteins orally. (IMAGE IS COURTESY OF UNIGENE LABORATORIES)

ADVANSTAR



2011 Advanstar Communications Inc.. Permission granted for up to 5 copies. All rights reserved.

You may forward this article or get additional permissions by typing http://license.icopyright.net/3.7458?icx_id=742139 into any web browser. Advanstar Communications Inc. and Pharmaceutical Technology logos are registered trademarks of Advanstar Communications Inc. The iCopyright logo is a registered trademark of iCopyright, Inc.